

BIOLOGICAL BULLETIN

THE RELATION BETWEEN THE RESPONSES BY *AMÆBA* TO MECHANICAL SHOCK AND TO SUDDEN ILLUMINATION.¹

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The reactions induced in *Amæba* by mechanical shock are remarkably like those brought about by sudden illumination (Folger, '25, '26). In both cases the response consists of a cessation of movement, the animal remaining inactive for a short time, then resuming locomotion. In both cases a short period intervenes between the application of the stimulus and the response, and this period, the reaction-time, varies directly with the magnitude of the stimulation, becoming shorter as the latter increases; while the time during which the amœba is inactive, the period of quiescence, likewise dependent on the magnitude of the stimulus, becomes longer with increase of the latter. Moreover, in both instances a certain amount of time must elapse after a stimulus has been applied before the amœba will respond to a second stimulus. Thus, an exposure to light of sufficient duration to bring about a response must be followed by an absence of light or at least by a lowered intensity for a certain amount of time before the amœba will again respond to an increase in illumination. This occurrence of a period during which the amœba is apparently reverting to the condition it was in before stimulation, which I have called the period of recovery,² and the fact that a recovery occurs

¹ Contribution from the Zoölogical Laboratory of the University of Michigan.

² The term recovery has been used here simply because it is descriptive; refractory period has been employed and in the case of light dark adaptation. Recovery should not be confused with the resumption of protoplasmic flow by the amœba. The animal may have resumed locomotion and be moving



after a mechanical shock as well as after sudden illumination, is of value and has been used, as we shall see presently, for a study of the relationship which exists between the reactions caused by mechanical shock and those brought about by light.

In a study of the influence of light on *Amæba* (Folger, '25) the question occurred: since the animal must recover from the effects of a sudden illumination before it will again respond to sudden illumination, must it also recover from the effects of a mechanical shock before it will respond to sudden illumination? The answer was clearly an affirmative one. In many instances the mechanical shock entirely inhibited a response to a sudden increase in luminous intensity, when the latter was applied shortly afterward. In these instances, however, the shock was brought about by moving the coverslip with the tip of a lead pencil, a method which, while it did produce undoubted results, is extremely crude. As a better means for controlling mechanical shock has since been used, the experiment dealing with the effect of this stimulus on the response to sudden illumination has been repeated and the results are presented in the following pages.

Moreover, if a mechanical shock preceding sudden illumination influences the response to the latter, the question also arises, is the reverse true? Does sudden illumination affect the response to mechanical shock? The results of experiments designed to answer this question are also set forth in this paper.

MATERIALS AND METHODS.

Specimens of *Amæba proteus* were used in the experiments, raised in small glass vessels containing a culture solution formed by adding raw hay to distilled water. Large and active individuals were selected for experimentation.

Two sets of apparatus were employed, one to bring about sudden illumination, the other to cause a mechanical shock. The latter was obtained by allowing a copper wire, weighing about 300 mg., to drop through a glass tube, 68 cm. in length, which was supported by a stand and clamps in such a manner that the weight rapidly and still not have "recovered" from the effects of stimulation as indicated by the fact that it fails to respond to a second exposure to illumination.

struck one end of the slide containing the amœba. Light was procured from a 1,000-watt, 112-volt, cylindrical Mazda stereopticon lamp, and flashed upon the amœba by means of the plane mirror of the microscope, set at an angle of 45 degrees. An intensity of about 16,000 meter candles was employed. To observe the organism when it was not illuminated by the strong light use was made of a Spencer miniature substage lamp.

The amœba to be experimented on was placed in a drop of water on a glass slide, within a ring of vaseline, and beneath a thin coverslip which was supported on one side by a small glass rod. The microscope was so arranged that when the amœba was in position under the lens, light could be flashed upon it or a mechanical shock applied at the will of the investigator, and the effect of one stimulus upon the other noted.

EXPERIMENTAL RESULTS.

Table I. illustrates the effect of mechanical shock upon the re-

TABLE I.
ILLUSTRATING THE EFFECT OF MECHANICAL SHOCK UPON THE RESPONSE TO
SUDDEN ILLUMINATION.

In each trial the amœba was exposed to strong illumination, and in every other one it was subjected to a mechanical shock before being illuminated. Three minutes were allowed between tests.

	Number of Trials.	No Reactions.
Sudden illumination alone.....	5	0
Sudden illumination following shortly after a mechanical shock	6	4

sponse to light. The experiment from which the data in this table were derived consisted of a number of tests, in each of which the amœba was exposed to strong illumination, and in every other one of which it was subjected to a mechanical shock before being illuminated. This shock was of sufficient magnitude to cause a cessation of movement, and the animal was exposed to light immediately on the resumption of flow. An interval of three minutes between tests permitted a recovery from the effects of previous stimulation. As shown in the table the amœba failed to respond to light 4 times out of 6 trials when a mechanical shock

preceded the exposure to illumination, while it responded 5 times out of 5 trials when previous stimulation by mechanical shock was lacking. These results are entirely in accord with previous observations and leave no doubt that mechanical shock does affect the response to light.

Table II. is the record of an experiment in which the procedure

TABLE II.

SHOWING THE EFFECT OF SUDDEN ILLUMINATION UPON THE RESPONSE TO MECHANICAL SHOCK.

In each trial the animal was subjected to a mechanical shock, and in every other one it was illuminated before being exposed to shock. Three minutes were allowed between tests.

Individual No.		Number of Trials.	No Reactions.
1	Mechanical shock alone	5	0
	Mechanical shock following immediately after sudden illumination	6	3
2	Mechanical shock alone	6	1
	Mechanical shock following immediately after sudden illumination	8	4
3	Mechanical shock alone	6	1
	Mechanical shock following immediately after sudden illumination	6	5
4	Mechanical shock alone	4	0
	Mechanical shock following immediately after sudden illumination	6	4
Totals	Mechanical shock alone	21	2
	Mechanical shock following immediately after sudden illumination	26	16

in the experiment just described was reversed. Here tests in which the animal was stimulated by illumination and immediately on the resumption of movement given a mechanical shock alternated with tests in which a mechanical shock alone was used as the stimulating agent. As shown in the table, individual No. 1 reacted 5 times out of 5 trials when stimulated by mechanical shock alone, while it reacted to the same stimulus only 3 times out of 6 trials when the mechanical shock was preceded by ex-

posure to light. Individual No. 2 reacted 5 times out of 6 trials when subjected to mechanical shock alone, and only 4 times out of 8 trials when the mechanical shock was preceded by illumination. Altogether, the 4 animals used in the experiment responded to mechanical shock 19 times out of 21 trials when this stimulus did not follow an exposure to light, while they failed to respond to it 16 times out of 26 trials when it did follow illumination, thus indicating that exposure to light does influence the response to mechanical shock. Even more convincing, however, are the results of an experiment to be described in the next paragraph.

It has already been shown (Folger, '26) that the length of time that an amoeba remains inactive after stimulation by a mechanical shock is markedly affected by the length of time that has elapsed since a previous mechanical shock, and that, within limits, this quiescent period increases with increase in the length of time since the previous stimulation. Thus, the period of quiescence resulting from a mechanical shock which follows 30 seconds after a preceding similar shock is not likely to be nearly so long as that brought about by a shock following after an interval of 60 seconds. Table III. records an experiment in which somewhat similar re-

TABLE III.
SHOWING THE EFFECT OF LIGHT ON MECHANICAL SHOCK.

In each test the amoeba was first exposed to light and then allowed to recover from the effects of this stimulus for the time indicated in the table, after which it was subjected to a mechanical shock. The period of quiescence, recorded in the last column, consists of the time during which the amoeba was inactive after it had been subjected to a mechanical shock.

Time Allowed for Recovery Between Sudden Illumination and Mechanical Shock (Sec.).	Reactions.	No Reactions.	Average Period of Quiescence (Sec.).
15.....	2	2	4.5
20.....	3	0	5.8
25.....	4	0	9.1
35.....	2	0	20.0

sults were obtained, but in which the organism was first stimulated by light and then by mechanical shock. From the table it is seen that the amoeba reacted only twice out of 4 trials when 15 seconds were allowed for recovery between stimulation by light and by

mechanical shock, and that the period of quiescence amounted to 4.5 seconds. Twenty seconds for recovery resulted in 3 reactions out of 3 trials, with an average period of quiescence of 5.8 seconds, 25 seconds for recovery resulted in 4 reactions out of 4 trials, with an average period of quiescence of 9.1 seconds, and 35 seconds for recovery resulted in 2 reactions out of 2 trials, with an average period of quiescence of 20 seconds.

Thus it appears that not only does sudden illumination affect the response of an amoeba to mechanical shock, but that it has precisely the same effect as another mechanical shock.

DISCUSSION.

A similarity of response to various kinds of stimuli has been noted in other organisms. Ewart ('03) has collected considerable data concerning protoplasmic streaming in plant cells, especially in the cells of *Chara* and *Nitella*. He and others have found that the cells react in a very characteristic way to various stimulating agents, the response consisting, just as in *Amoeba*, of a temporary cessation of movement. The best quantitative results were obtained by means of a mechanical shock, brought about by dropping a weight on the coverslip beneath which the streaming cells had been placed. By using weights of various sizes a gradation in the magnitude of the shock was possible. Ewart discovered that streaming did not stop immediately on application of the stimulus, but after the intervention of a reaction-time, which was longer after a light shock than after one of greater magnitude and which in the event of a sub-minimal stimulus might amount to 7 or 8 seconds. He found, furthermore, that the time during which movement remained suspended likewise depended on the magnitude of the shock, streaming recommencing much sooner after a slight shock than after a heavy one. Various other stimulating agents, including light, heat, electricity, and change in concentration of the surrounding medium, gave very similar results, although in no instance was so accurate a quantitative measurement obtained as with mechanical shock. Ewart also noted that the application of one kind of stimulus may tend to inhibit the response to another kind.

The refractory period used as a basis for the experiments presented in this paper, during which recovery from the effects of previous stimulation occurs, has long been known, especially when light has been used as the stimulating agent. In this case the term dark adaptation has been used. An animal must be dark-adapted before it will respond to illumination. This necessity for dark-adaptation has led to an explanation of the response to light which involves the presence in the organism of a substance capable of a reversible photochemical reaction (Mast, '07; Hecht, '18). The reaction is thought of as being initiated by the conversion of a photosensitive substance into its precursors, and dark-adaptation as the reforming of this substance from the precursors.

As we have just seen, however, recovery from the effects of a mechanical shock may be necessary before a response to sudden illumination is possible, and it would therefore seem that the same processes are involved in the refractory periods which follow the two types of stimuli. If so, since a photochemical reaction cannot occur in the case of mechanical shock, it is evident that in *Amœba*, at least, dark-adaptation must consist of something besides the reforming of a photosensitive substance from its precursors.

SUMMARY.

1. *Amœba* responds both to mechanical shock and to sudden illumination by a cessation of movement, which does not take place immediately on stimulation, but after a considerable reaction-time.

2. In both cases the reaction-time depends upon the magnitude of the stimulus, becoming longer as the intensity of the stimulating agent increases.

3. In both instances the time during which the amœba is inactive also depends upon the magnitude of the stimulus, becoming longer as the latter increases.

4. After an amœba has been exposed to light it is necessary that a certain interval of time elapse before it will again respond to sudden illumination. Likewise, after a mechanical shock the amœba must be allowed time for recovery before it will respond to a second shock.

5. After a response to light time must be allowed for recovery before the amoeba will react to mechanical shock and vice versa.

6. The effect of one kind of stimulus upon a response to another kind leads one to infer that the processes occurring during the refractory periods following the reactions caused by mechanical shock and by sudden illumination are basically the same.

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